

## SEQUENCE LISTING

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NEW ENGLAND BIOLABS, INC.  
BOSTON BIOMEDICAL RESEARCH INSTITUTE

<120> METHOD FOR GENERATING SPLIT, NON-TRANSFERABLE GENES  
THAT ARE ABLE TO EXPRESS AN ACTIVE PROTEIN PRODUCT

<130> NEB-163-PCT

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<150> 60/135,677  
<151> 1999-05-24

<160> 134

<170> PatentIn Ver. 2.0

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Val His Val Asp Ile Asp Pro Ala Glu Ile Gly Lys Asn Lys Gln Pro  
35 40 45

His Val Ser Ile Cys Ala Asp Val Lys Leu Ala Leu Gln Gly Met Asn  
50 55 60

Ala Leu Leu Glu Gly Ser Thr Ser Lys Lys Ser Phe Asp Phe Gly Ser  
65 70 75 80

Trp Asn Asp Glu Leu Asp Gln Gln Lys Arg Glu Phe Pro Leu Gly Tyr  
85 90 95

Lys Thr Ser Asn Glu Glu Ile Gln Pro Gln Tyr Ala Ile Gln Val Leu  
100 105 110

Asp Glu Leu Thr Lys Gly Glu Ala Ile Ile Gly Thr Gly Val Gly Gln  
115 120 125

His Gln Met Trp Ala Ala Gln Tyr Tyr Thr Tyr Lys Arg Pro Arg Gln  
130 135 140

Trp Leu Ser Ser Ala Gly Leu Gly Ala Met Gly Phe Gly Leu Pro Ala  
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Ala Ala Gly Ala Ser Val Ala Asn Pro Gly Val Thr Val Val Asp Ile  
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Asp Gly

<210> 43

<211> 179

<212> PRT

<213> Escherichia coli

<400> 43

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Ile Val His Ile Asp Ile Asp Ser Ala Glu Ile Gly Lys Asn Lys Gln	35	40	45
Pro His Val Ser Ile Cys Ala Asp Ile Lys Leu Ala Leu Gln Gly Leu	50	55	60
Asn Ser Ile Leu Glu Ser Lys Glu Gly Lys Leu Lys Leu Asp Phe Ser	65	70	75
Ala Trp Arg Gln Glu Leu Thr Glu Gln Lys Val Lys His Pro Leu Asn	85	90	95
Phe Lys Thr Phe Gly Asp Ala Ile Pro Pro Gln Tyr Ala Ile Gln Val	100	105	110
Leu Asp Glu Leu Thr Asn Gly Asn Ala Ile Ile Ser Thr Gly Val Gly	115	120	125
Gln His Gln Met Trp Ala Ala Gln Tyr Tyr Lys Tyr Arg Lys Pro Arg	130	135	140
Gln Trp Leu Thr Ser Gly Gly Leu Gly Ala Met Gly Phe Gly Leu Pro	145	150	155
Ala Ala Ile Gly Ala Ala Val Gly Arg Pro Asp Glu Val Val Val Asp	165	170	175
Ile Asp Gly			

<210> 44

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<213> Escherichia coli

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Tyr Ala Val Asp Ser Ser Asp Leu Leu Leu Ala Phe Gly Val Arg Phe	1	5	10	15
Asp Asp Arg Val Thr Gly Lys Leu Glu Ala Phe Ala Ser Arg Ala Lys	20	25	30	
Ile Val His Ile Asp Ile Asp Ser Ala Glu Ile Gly Lys Asn Lys Gln	35	40	45	
Pro His Val Ser Ile Cys Ala Asp Ile Lys Leu Ala Leu Gln Gly Leu	50	55	60	
Asn Ser Ile Leu Glu Ser Lys Glu Gly Lys Leu Lys Leu Asp Phe Ser	65	70	75	80

Ala Trp Arg Gln Glu Leu Thr Val Gln Lys Val Lys Tyr Pro Leu Asn  
85 90 95

Phe Lys Thr Phe Gly Asp Ala Ile Pro Pro Gln Tyr Ala Ile Gln Val  
100 105 110

Leu Asp Glu Leu Thr Asn Gly Ser Ala Ile Ile Ser Thr Gly Val Gly  
115 120 125

Gln His Gln Met Trp Ala Ala Gln Tyr Tyr Lys Tyr Arg Lys Pro Arg  
130 135 140

Gln Trp Leu Thr Ser Gly Gly Leu Gly Ala Met Gly Phe Gly Leu Pro  
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Ala Ala Ile Gly Ala Ala Val Gly Arg Pro Asp Glu Val Val Val Asp  
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Ile Asp Gly

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<213> Escherichia coli

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Val Leu His Ile Asp Ile Asp Pro Thr Ser Ile Ser Lys Thr Val Thr  
35 40 45

Ala Asp Ile Pro Ile Val Gly Asp Ala Arg Gln Val Leu Glu Gln Met  
50 55 60

Leu Glu Leu Leu Ser Gln Glu Ser Ala His Gln Pro Leu Asp Glu Ile  
65 70 75 80

Arg Asp Trp Trp Gln Gln Ile Glu Gln Trp Arg Ala Arg Gln Cys Leu  
85 90 95

Lys Tyr Asp Thr His Ser Glu Lys Ile Lys Pro Gln Ala Val Ile Glu  
100 105 110

Thr Leu Trp Arg Leu Thr Lys Gly Asp Ala Tyr Val Thr Ser Asp Val  
115 120 125

Gly Gln His Gln Met Phe Ala Ala Leu Tyr Tyr Pro Phe Asp Lys Pro  
130 135 140

Arg Arg Trp Ile Asn Ser Gly Gly Leu Gly Thr Met Gly Phe Gly Leu  
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Cys Val Thr Gly  
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<212> PRT

<213> Escherichia coli

<400> 46

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Val Ile His Met Asp Ile Asp Pro Ala Glu Met Asn Lys Leu Arg Gln  
35 40 45

Ala His Val Ala Leu Gln Gly Asp Leu Asn Ala Leu Leu Pro Ala Leu  
50 55 60

Gln Gln Pro Leu Asn Gln Cys Asp Trp Gln Gln His Cys Ala Gln Leu  
65 70 75 80

Arg Asp Glu His Ser Trp Arg Tyr Asp His Pro Gly Asp Ala Ile Tyr  
85 90 95

Ala Pro Leu Leu Leu Lys Gln Leu Ser Asp Arg Lys Pro Ala Asp Cys  
100 105 110

Val Val Thr Thr Asp Val Gly Gln His Gln Met Trp Ala Ala Gln His  
115 120 125

Ile Ala His Thr Arg Pro Glu Asn Phe Ile Thr Ser Ser Gly Leu Gly  
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Pro Asn Asp Thr Val Val Cys Ile Ser Gly  
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ends of the Tn7 transposon

<400> 86  
Cys Leu Asn Thr Leu  
1 5

<210> 87  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 87  
Val Phe Lys Gln Pro  
1 5

<210> 88  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 88  
Cys Leu Asn Ser Met  
1 5

<210> 89  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 89  
Cys Leu Asn Asn Tyr  
1 5

<210> 90  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 90  
Cys Leu Asn Met Ala  
1 5



<210> 91  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 91  
Val Phe Lys His Lys  
1 5

<210> 92  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 92  
Cys Leu Asn Thr Lys  
1 5

<210> 93  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 93  
Cys Leu Asn Lys Asp  
1 5

<210> 94  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 94  
Met Phe Lys Gln Ile  
1 5

<210> 95  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 95  
Cys Leu Asn Ile Ile  
1 5

<210> 96  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 96  
Leu Phe Lys His Glu  
1 5

<210> 97  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 97  
Val Phe Lys His Phe  
1 5

<210> 98  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 98  
Cys Leu Asn Ser Val  
1 5

<210> 99  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 99  
Val Phe Lys Gln Ile  
1 5

<210> 100  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 100  
Met Phe Lys Gln Ala  
1 5

<210> 101  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 101  
Leu Phe Lys His His  
1 5

<210> 102  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 102  
Leu Phe Lys His Gln  
1 5

<210> 103  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 103  
Met Phe Lys His Val  
1 5

<210> 104  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 104  
Val Phe Lys Gln Lys  
1 5

<210> 105  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 105  
Leu Phe Lys Gln Gln  
1 5

<210> 106  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 106  
Leu Phe Lys His Ser  
1 5

<210> 107  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 107  
Cys Leu Asn Thr Gly  
1 5

<210> 108  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 108  
Cys Leu Asn Ser Arg  
1 5

<210> 109  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 109  
Val Phe Lys His Leu  
1 5

<210> 110  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 110  
Cys Leu Asn Asn Ile  
1 5

<210> 111  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 111  
Leu Phe Lys His Gln  
1 5

<210> 112  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 112  
Cys Leu Asn Lys His  
1 5

<210> 113  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 113  
Met Phe Lys Gln Tyr  
1 5

<210> 114  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 114  
Cys Leu Asn Lys Gln  
1 5

<210> 115  
<211> 5  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 115  
Cys Leu Asn Met Ser  
1 5

<210> 116  
<211> 7  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 116  
Leu Cys Leu Asn Ile Leu Ala  
1 5

<210> 117  
<211> 7  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 117  
Asn Cys Leu Asn Ile Asn Ala  
1 5

<210> 118  
<211> 7  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 118  
Leu Met Phe Lys His Leu Ser  
1 5

<210> 119  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 119  
Thr Leu Phe Lys His Thr Arg  
1 5

<210> 120  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 120  
Lys Val Phe Lys Gln Lys Glu  
1 5

<210> 121  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 121  
His Leu Val Phe Lys His Leu  
1 5

<210> 122  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 122  
Leu Cys Leu Asn Thr Leu Leu  
1 5



<210> 123  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 123  
Leu Cys Leu Asn Asn Leu Val  
1 5

<210> 124  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 124  
Glu Val Phe Lys His Glu Gly  
1 5

<210> 125  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 125  
Lys Val Phe Lys Gln Lys Gly  
1 5

<210> 126  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 126  
Thr Cys Leu Asn Thr Thr Ile  
1 5

<210> 127  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 127  
Met Cys Leu Asn Asn Met Asn  
1 5

<210> 128  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 128  
Leu Leu Phe Lys Gln Leu Arg  
1 5

<210> 129  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 129  
Arg Cys Leu Asn Asn Arg Leu  
1 5

<210> 130  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 130  
Met Val Phe Lys Gln Met Ala  
1 5

<210> 131  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 131  
Ala Met Phe Lys Gln Ala Thr  
1 5

<210> 132  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 132  
Leu Val Phe Lys His Leu Asp  
1 5

<210> 133  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 133  
Lys Met Phe Lys Gln Lys Thr  
1 5

<210> 134  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: based on the  
ends of the Tn7 transposon

<400> 134  
Tyr Cys Leu Asn Asn Tyr Phe  
1 5